

AMENDMENT AND RESPONSE

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Applicant(s): Nancy D. HANSON et al.
Serial No.: 09/814,257
Filed: 21 March 2001
For: PRIMERS FOR USE IN DETECTING BETA-LACTAMASES

Amendments to the Claims

This listing of claims replaces all prior versions, and listings, of claims in the above-identified application:

- 1-11. (canceled)
12. (previously presented) A primer selected from the group of:
5' - CGT CGC TCA CCA TAT CTC CC - 3' (SEQ ID NO:34);
5' - CCT CTC GTG CTT TAG ACC CG - 3' (SEQ ID NO:35); and full-length complements thereof.
13. (previously presented) A primer selected from the group of:
5' - CGC TGG GAA ACC TAT TCG G - 3' (SEQ ID NO:36);
5' - CTG CCA TCC AGT TTC TTC GGG - 3' (SEQ ID NO:37); and full-length complements thereof.
14. (previously presented) A primer selected from the group of:
5' - GGT GGC ATT GAC AAA TTC TGG - 3' (SEQ ID NO:38);
5' - CCC ACC ATG CGA CAC CAG - 3' (SEQ ID NO:39); and full-length complements thereof.
15. (previously presented) A primer selected from the group of:
5' - TGT GCA ACG CAA ATG GCA C - 3' (SEQ ID NO:40);
5' - CGA CCC CAA GTT TCC TGT AAG TG - 3' (SEQ ID NO:41); and full-length complements thereof.

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16. (previously presented) A primer selected from the group of:

5' - AGG CAC GAT AGT TGT GGC AGA C - 3' (SEQ ID NO:42);

5' - CAC TCA ACC CAT CCT ACC CAC C - 3' (SEQ ID NO:43); and full-length complements thereof.

17.-50. (canceled)

51. (previously presented) A diagnostic kit for detecting an OXA family beta-lactamase which comprises packaging, containing, separately packaged:

(a) at least one primer pair capable of hybridizing to beta-lactamase nucleic acid of interest;

(b) a positive and negative control; and

(c) a protocol for identification of the beta-lactamase nucleic acid of interest;

wherein the primers are selected from the group consisting

of:

5' - CGT CGC TCA CCA TAT CTC CC - 3' (SEQ ID NO:34);

5' - CCT CTC GTG CTT TAG ACC CG - 3' (SEQ ID NO:35);

5' - CGC TGG GAA ACC TAT TCG G - 3' (SEQ ID NO:36);

5' - CTG CCA TCC AGT TTC TTC GGG - 3' (SEQ ID NO:37);

5' - GGT GGC ATT GAC AAA TTC TGG - 3' (SEQ ID NO:38);

5' - CCC ACC ATG CGA CAC CAG - 3' (SEQ ID NO:39);

5' - TGT GCA ACG CAA ATG GCA C - 3' (SEQ ID NO:40);

5' - CGA CCC CAA GTT TCC TGT AAG TG - 3' (SEQ ID NO:41);

5' - AGG CAC GAT AGT TGT GGC AGA C - 3' (SEQ ID NO:42);

5' - CAC TCA ACC CAT CCT ACC CAC C - 3' (SEQ ID NO:43); and full-length complements thereof.

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52. (currently amended) A method for identifying a beta-lactamase in a clinical sample, the method comprising:

providing a pair of oligonucleotide primers specific for nucleic acid characteristic of the OXA family of beta-lactamase enzymes, wherein one primer of the pair is complementary to at least a portion of the beta-lactamase nucleic acid in the sense strand and the other primer of the pair is complementary to at least a portion of the beta-lactamase nucleic acid in the antisense strand;

annealing the primers to the beta-lactamase nucleic acid;

simultaneously extending the annealed primers from a 3' terminus of each primer to synthesize an extension product that is complementary to the nucleic acid strands annealed to each primer wherein each extension product after separation from the beta-lactamase nucleic acid serves as a template for the synthesis of an extension product using a primer that is complementary to said each extension product;

separating the amplified products; and

analyzing the separated amplified products for a region characteristic of the beta-lactamase;

wherein the primers are selected from the group consisting of:

5' - CGT CGC TCA CCA TAT CTC CC - 3' (SEQ ID NO:34);

5' - CCT CTC GTG CTT TAG ACC CG - 3' (SEQ ID NO:35);

5' - CGC TGG GAA ACC TAT TCG G - 3' (SEQ ID NO:36);

5' - CTG CCA TCC AGT TTC TTC GGG - 3' (SEQ ID NO:37);

5' - GGT GGC ATT GAC AAA TTC TGG - 3' (SEQ ID NO:38);

5' - CCC ACC ATG CGA CAC CAG - 3' (SEQ ID NO:39);

5' - TGT GCA ACG CAA ATG GCA C - 3' (SEQ ID NO:40);

5' - CGA CCC CAA GTT TCC TGT AAG TG - 3' (SEQ ID NO:41);

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5' - AGG CAC GAT AGT TGT GGC AGA C - 3' (SEQ ID NO:42);

5' - CAC TCA ACC CAT CCT ACC CAC C - 3' (SEQ ID NO:43); and full-length complements thereof.

53. (currently amended) A method for identifying a beta-lactamase in a clinical sample, the method comprising:

providing a pair of oligonucleotide primers specific for nucleic acid characteristic of the OXA family of beta-lactamase enzymes, wherein one primer of the pair is complementary to at least a portion of the beta-lactamase nucleic acid in the sense strand and the other primer of the pair is complementary to at least a portion of the beta-lactamase nucleic acid in the antisense strand;

annealing the primers to the beta-lactamase nucleic acid;

simultaneously extending the annealed primers from a 3' terminus of each primer to synthesize an extension product that is complementary to the nucleic acid strands annealed to each primer wherein each extension product after separation from the beta-lactamase nucleic acid serves as a template for the synthesis of an extension product using a primer that is complementary to said each extension product;

separating the amplified products; and

analyzing the separated amplified products for a region characteristic of the at least one beta-lactamase selected from the group consisting of OXA-1, OXA-2, OXA-3, OXA-5, OXA-6, OXA-7, OXA-9, OXA-10, OXA-11, OXA-12, OXA-13, OXA-14, OXA-15, and combinations thereof;

wherein when the oligonucleotide primers are specific for the OXA family beta-lactamase enzyme designated as OXA-1, the primers are selected from the group of:

5' - TGT GCA ACG CAA ATG GCA C - 3' (SEQ ID NO:40);

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5' - CGA CCC CAA GTT TCC TGT AAG TG - 3' (SEQ ID NO:41); and full-length complements thereof;

wherein when the oligonucleotide primers are specific for the OXA family beta-lactamase enzymes designated as OXA-5, 6, 7, 10, 11, 13, or 14, the primers are selected from the group of:

5' - GGT GGC ATT GAC AAA TTC TGG - 3' (SEQ ID NO:38);

5' - CCC ACC ATG CGA CAC CAG - 3' (SEQ ID NO:39); and full-length complements thereof;

wherein when the oligonucleotide primers are specific for the OXA family beta-lactamase enzyme designated as OXA-9, the primers are selected from the group of;

5' - CGT CGC TCA CCA TAT CTC CC - 3' (SEQ ID NO:34);

5' - CCT CTC GTG CTT TAG ACC CG - 3' (SEQ ID NO:35); and full-length complements thereof;

wherein when the oligonucleotide primers are specific for the OXA family beta-lactamase enzyme designated as OXA-12, the primers are selected from the group of:

5' - CGC TGG GAA ACC TAT TCG G - 3' (SEQ ID NO:36);

5' - CTG CCA TCC AGT TTC TTC GGG - 3' (SEQ ID NO:37); and full-length complements thereof;

and wherein when the oligonucleotide primers are specific for the OXA family beta-lactamase enzyme designated as OXA-2, 3, or 15, the primers are selected from the group of:

5' - AGG CAC GAT AGT TGT GGC AGA C - 3' (SEQ ID NO:42);

5' - CAC TCA ACC CAT CCT ACC CAC C - 3' (SEQ ID NO:43); and full-length complements thereof.

54. (currently amended) A method for identifying a beta-lactamase in a clinical sample, the method comprising:

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providing a pair of oligonucleotide primers specific for nucleic acid encoding an OXA family beta-lactamase enzyme, wherein the enzyme is found in a Gram-negative bacterium selected from the group of *Enterbacter cloacae*, *Citrobacter freundii*, *Serratia marcescens*, *Providencia spp.*, *Proteus mirabilis*, *Yersinia enterocolitica*, and combinations thereof, wherein one primer of the pair is complementary to at least a portion of the beta-lactamase nucleic acid in the sense strand and the other primer of the pair is complementary to at least a portion of the beta-lactamase nucleic acid in the antisense strand;

annealing the primers to the beta-lactamase nucleic acid;

simultaneously extending the annealed primers from a 3' terminus of each primer to synthesize an extension product that is complementary to the nucleic acid strands annealed to each primer wherein each extension product after separation from the beta-lactamase nucleic acid serves as a template for the synthesis of an extension product using a primer that is complementary to said each extension product;

separating the amplified products; and

analyzing the separated amplified products for a region characteristic of a beta-lactamase;

wherein the primers are selected from the group consisting of:

5' - CGT CGC TCA CCA TAT CTC CC - 3' (SEQ ID NO:34);

5' - CCT CTC GTG CTT TAG ACC CG - 3' (SEQ ID NO:35);

5' - CGC TGG GAA ACC TAT TCG G - 3' (SEQ ID NO:36);

5' - CTG CCA TCC AGT TTC TTC GGG - 3' (SEQ ID NO:37);

5' - GGT GGC ATT GAC AAA TTC TGG - 3' (SEQ ID NO:38);

5' - CCC ACC ATG CGA CAC CAG - 3' (SEQ ID NO:39);

5' - TGT GCA ACG CAA ATG GCA C - 3' (SEQ ID NO:40);

5' - CGA CCC CAA GTT TCC TGT AAG TG - 3' (SEQ ID NO:41);

5' - AGG CAC GAT AGT TGT GGC AGA C - 3' (SEQ ID NO:42);